RESISTANCE TO BARLEY LEAF RUST (*PUCCINIA ANOMALA* ROST.).*

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Introduction.

Although considerable work has been done on the genetics of barley in the United States, little is known of the way in which resistance to leaf rust (*Puccinia anomala*) is inherited. Studies have been made of the reactions of barley varieties to this disease, but, as far as the writers are aware, it is not known whether an allelic series of genes controls the different types of resistance that have been found or whether several loci are involved. In this country the only reported work dealing with the genetics of resistance to barley leaf rust was done by Waterhouse (1927), and he found simple monofactorial segregations in crosses between resistant and susceptible types.

The disease is not of economic importance in New South Wales. It occurs commonly along the eastern coast but never reaches epiphytotic proportions in the main barley growing areas of the State. Hence this study was not connected with any breeding project but was undertaken to determine whether the occurrence of several factors for resistance to barley mildew, *Erysiphe graminis Hordei*, as found by Briggs and his colleagues in California, was paralleled by a similar group for resistance to *P. anomala*. This paper presents our preliminary results.

Varietal Reactions.

Throughout the work the same rust culture was used as that previously reported upon (Waterhouse, 1927). At the time when this latter work was done no set of differential varieties had been established for the identification of physiological races of *P. anomala*. In 1939 d'Oliveira published a list of eleven varieties which served to distinguish between the 30 races that had been isolated in Europe up till that time. Through the courtesy of Dr. d'Oliveira, who kindly supplied us with seed, we have been able to compare certain Australian cultures with those occurring elsewhere.

The work that was done earlier suggested that more than one race of *P. anomala* is present here. Waterhouse (1927) found *Hordeum murinum* (*Hordeum leporinum*) to be resistant. Later (1929) he listed this same species as susceptible, and the logical explanation is that two different physiological races were involved when these tests were made, although variation in the host plant could have caused a similar result. We have tested collections of this organism on the above grass and so far have been unable to detect any differences between them. *Hordeum leporinum* has proved immune in all tests.

When d'Oliveira's varieties were inoculated the following reactions were obtained with rust collections from Werribee, Victoria; Lawes, Queensland; Wingen and Muswellbrook, N.S.W. The figures represent the usual designations used in cereal leaf rust studies and are comparable with d'Oliveira's own descriptions (see Table 1).

In some of these varieties it was apparent that the seed was not pure for its reactions to these collections of rust. *H. vulgare speciale*, Aegyptische 4-zeilige and Oderbrucker were outstanding in this regard. There was clear evidence of both resistant

* This paper is the result of research work carried out in the Faculty of Agriculture, the University of Sydney.

TABLE 1.

Variety.	S.U. Accession Number.	Rust Reaction.
Brustedt's Schladener	B 244	0, 1
Hordeum vulgare speciale	B 245	0, 1, 2, 4
Friedrichswerther Berg Wintergerste	B 246	4
Australische Recka	B 247	0, 1
Lichtis Lechtaler	B 248	4
Samaria 4–zeilige	B 249	4
Hordeum vulgare pallidum	B 250	4
Aegyptische 4–zeilige sommergerste	B 251	0, 1, 2, 4
Quinn C.I. 1024	B 252	0, 1, 2
Bolivia C.I. 1257	B 253	0, 1
Oderbrucker C.I. 940	B 254	0, 1

and susceptible plants in the pots of these varieties. *H. vulgare speciale* and Aegyptische 4-zeilige had approximately the same number of resistant and susceptible plants and they are accordingly designated 0, 1, 2, 4. Oderbrucker, although it contained a mixture of types, was in the main susceptible. Seed of Oderbrucker C.I. 940 which we had on hand, however, was uniformly resistant to rust and consequently the reaction has been left as 0, 1, although the seed from Portugal did not give precisely this reaction. The four collections that were compared on this series of varieties were also studied on other varieties listed by d'Oliveira. The reactions were (see Table 2):

TABLE 2.

Variety.	S.U. Accession Number.	Rust Reaction.
Hordeum hexastichum eurylepis	B 255	4
,, ,, *recens*	B 256	0, 1, 2
Ackermann's Bavaria	B 257	4
Featherston C.I. 1120	B 258	0, 1
Malting C.I. 1129	B 259	0, 1
Hanna C.I. 906	B 260	4
Juliaca C.I. 1114	B 261	0, 1

The above results, together with those given earlier, indicate that these collections of rust represent a physiological race unlike any of the 30 reported from Europe. In addition they have been found to differ from the races 1 and 2 given by Mains (1932) for U.S.A. In order to compare the race to which these collections belong, with races from overseas, we have tested all the barley varieties that have been added to the University accession lists since the time of Waterhouse's report in 1927. Unfortunately no field results are available, but the following classification will serve to indicate seedling reactions to our collections of leaf rust. Certain varieties listed above are given for comparison in Table 3.

Among the varieties listed as resistant there is a considerable amount of variation, but in all cases the reaction is one where much flecking results and no fully susceptible pustules are formed. Those varieties given as moderately resistant can show quite a considerable development of rust. On the same leaf, however, there may be intermingled flecks (;) and 2– reactions and in isolated cases a pustule will develop which is indistinguishable from a fully susceptible type. However, when one becomes familiar with the varieties and their rust reaction there is seldom any doubt about the group for any particular variety.

TABLE 3.

Resistant	S.U. Accession Number.	Moderately Resistant.	S.U. Accession Number.	Susceptible.	S.U. Accession Number.
Featherston	B 129	Abyssinian	B 158	Abyssinian	B 124
Oderbrucker C.I. 940	B 130	Tennessee Winter	B 161	Athos	B 125
Oderbrucker C.I. 957	B 131	Ben Beardless	B 162	Club Hybrid	B 126
Unnamed C.I. 1347	B 132	017	B 173	Guimalaya	B 127
Malting C.I. 1129	B 133	Kwan	B 174	Smooth Awn × Beard-	
Manchuria C.I. 2330	B 134	Weider	B 175	less × Reka	B 128
Hooded Spring C.I. 716	B 135	Arequipa C.I. 1256	B 188	Goldfoil	B 167
Horsford C.I. 507	B 136	Callas C.I. 240	B 138	Hanna	B 168
Horsford C.I. 877	B 137	Mecknos Moroc C.I. 1379	B 139	Bel. 2971	B 171
Success	B 146	Peruvian C.I. 935	B 140	Peatland	B 176
Pearl	B 148	Quinn C.I. 1024	B 141	Nigrinudum	B 178
Alberta Beardless	B 189	Bolivia C.I. 1257	B 142	Compana	B 186
Glabron C.I. 4577	B 195	Unnamed C.I. 2329	B 143	Atlas × Vaughn	B 187
Colsess IV	B 212	Juliaca C.I. 1114	B 144	Californian Mariout	
Colsess V	B 213	Deputy	B 145	C.I. 3615	B 190
Afghanistan	B 221	Argentine	B 149	Chevron C.I. 1111	B 191
Schladener	B 242	O.A.C. 7	B 150	Charlottetown 80	B 192
		Karru	B 151	Comfort C.I. 2488	B 193
		Algerian	B 152	Golden Pheasant	B 196
		Beldi	B 153	Leiorrinchum	B 197
		Coast	B 154	Newal	B 198
		Portuguese	B 155	Regal	B 199
		Smooth Awn × Reka	B 156	Velvet	B 203
		Sulu C.I. 1022	B 200	Vaughn	B 204
		Success	B 201	Wisconsin 38	B 205
		Trebi	B 202	Nigrinudum I	B 206
		Coast II	B 210	Nudideficiens	B 207
				Minn. 72–8	B 208
				Minn. 84–7	B 209
				Coast III	B 211
				Heitmann's Surprise	B 222
				Black Hulless	B 224
				Hanna	B 225
				Bolsheriki	B 227
				Barbless	B 230
				Male sterile	B 231
				Gymnospermum	B 233
				Engawnless	B 234
				Triple Bearded Mariout	B 235
				Trebi 4	B 237
				Egypt	B 238
				Austral	B 239
				Berg	B 240
				Samaria	B 243

INHERITANCE STUDIES.

Two of the varieties listed by Waterhouse (1927) have been mainly considered in this work. They are Minn. II 21.15 Smooth Awn × Manchuria and No. 22. Seedlings of No. 22 under glass-house conditions at Sydney are not as resistant to leaf rust as those of Minn. II 21.15. The reaction of the latter is almost always characterized by the sharpest flecks, that of No. 22 varies from place to place on the leaf from a fleck (;) to a 2– reaction. This range, for convenience, has been designated an X type. This difference in the resistant reaction to leaf rust suggested that the character might be controlled by a non-allelic series of genes, as has been found for different types of resistance to *P. graminis Tritici* in the Kenya varieties of *T. vulgare*. (Watson and Waterhouse, 1945.)

Both of the above barley varieties have been crossed with susceptible types and each has been found to possess a single major gene which differentiates its particular type of resistance from susceptibility. Smooth Awn × Manchuria and No. 22 were accordingly crossed together reciprocally, and as no difference was shown, both this cross and its reciprocal are considered together. The grains resulting from crossing were divided, some were grown at the Cowra Experiment Farm and others at the University of Sydney. These latter were tested as seedlings for their reaction to leaf rust and the reaction of Minn. II 21.15 was dominant. Seedlings of a small F_2 population of 101 plants were tested for leaf rust reaction in 1944 and later were transplanted to the field, the remaining F_2 grain was grown at Hawkesbury Agricultural College. Of the 101 F_2 plants that were inoculated, two escaped infection, 76 gave flecks, 16 gave an X reaction and 7 were fully susceptible. No rust developed on the F_2 plants in the field at Richmond on account of the drought that prevailed in 1944.

Both batches, the classified material from the University and the unclassified from Richmond, were grown in duplicate pots and tested with rust as F_3 progenies in 1945. Only 65 of the 101 tested F_2 plants produced progenies and the behaviour of these was as in Table 4.

TABLE 4.

F_3 Reaction.		F_2 Reaction.		
Breeding Result.	Type of Reaction.	;	X	4
Homozygous	;	13		
Segregating	;, X, 4	17		
Segregating	; and X	18		
Homozygous	X	1	3	
Segregating	X and 4	2	6	1
Segregating	; and 4	2		
Homozygous	4			2
Total	—	53	9	3

These figures, although not large, can be explained on the assumption that the two single factors possessed by these two varieties are not allelic and are inherited independently. On the basis of the reaction of the F_1 plants the resistance of Minn. II 21.15 is epistatic to that of No. 22. Of the 16 plants of the X type only 9 produced an F_3 progeny. If these plants carry the factor of No. 22 either in the heterozygous or homozygous condition, then one plant in three should be homozygous for the X reaction and two should segregate for the X reaction and complete susceptibility. The results observed with these nine progenies fit the expectancy well. According to this explanation the double recessive genotype which produced susceptible plants in F_2 should give homozygous susceptible plants in F_3. One progeny segregated for the X reaction and apparently this plant was wrongly classified in F_2.

On the suggestion of Dr. D. W. Robertson the single factor for resistance which is present in Minn. II 21.15 and which gives plants with sharp flecks, has been designated Pa_1. The one possessed by No. 22, which allows some rust to develop, is Pa_2. If these factors are independent and show the dominance relations given above, the expected phenotype in F_2 and its F_3 progeny would be as in Table 5.

The ratio of 76:16:7 which was obtained on 99 F_2 plants agrees reasonably well with the 12:3:1 which would be expected. Further data to support this explanation were obtained when rust reactions were observed on 170 additional F_3 progenies arising from plants which were grown at Richmond and were unclassified in the F_2 generation.

TABLE 5.

F_2 Genotype.	Expected F_2 Phenotype.	Reaction Type.	Expected F_3 Segregation.	Reaction Type.
Pa_1 Pa_1 Pa_2 Pa_2	Resistant.	;	Homozygous resistant.	;
Pa_1 Pa_1 Pa_2 pa_2 ..	,,	;	Homozygous resistant.	;
Pa_1 pa_1 Pa_2 Pa_2	,,	;	Segregating.	; and X
Pa_1 pa_1 Pa_2 pa_2 ..	,,	;	Segregating.	;, X, 4
Pa_1 Pa_1 pa_2 pa_2 ..	,,	;	Homozygous resistant.	;
Pa_1 pa_1 pa_2 pa_2 ..	,,	;	Segregating.	; and 4
pa_1 pa_1 Pa_2 Pa_2 ..	Moderately resistant.	X	Homozygous resistant	X
pa_1 pa_1 Pa_2 pa_2 ..	Moderately resistant.	X	Segregating.	X and 4
pa_1 pa_1 pa_2 pa_2 ..	Susceptible.	4	Homozygous susceptible.	4

When the two groups of 65 and 170 were pooled the classification resulted as shown in Table 6.

TABLE 6.

F_3 Reaction.	Reaction Type.	Expectancy in 16.	Observed.	Expected.
Homozygous resistant ..	;	4	57	58.7500
Segregating	; and X	2	39	29.3750
Segregating	; and 4	2	18	29.3750
Segregating all types ..	;, X, 4	4	55	58.7500
Homozygous	X	1	18	14.6875
Segregating	X and 4	2	35	29.3750
Homozygous susceptible ..	4	1	13	14.6875
Total		16	235	235·0000

When χ^2 is calculated the value of 9·868 is found to have a probability between 0·20 and 0·10. From these data then it appears that both varieties are resistant to leaf rust, one is highly resistant, the other moderately resistant, a single major factor is present in both parents but the factors are inherited independently in crosses between them. Their rust reaction and proposed genotype would be as follows:

Parent.	Rust Reaction.	
Smooth Awn × Manchuria	Resistant.	Pa_1 Pa_1 pa_2 pa_2
No. 22	Moderately resistant.	pa_1 pa_1 Pa_2 Pa_2

Association with Other Characters.

The linkage relations of these two genes are at present under investigation. A number of crosses have been made in this connection but the genes so far have been found to be inherited independently of all those other characters so far studied. In the cross studied here, for example, Minn. II 21.15 has smooth awns, short rachilla hairs and is susceptible to race 3 of powdery mildew (*Erysiphe graminis Hordei*); No. 22 has rough awns, long rachilla hairs and is moderately resistant to powdery mildew. In order to determine whether there was any association between characters the awn indices of roughness of both the parents and F_2 plants were found by the method described by Hayes et al. (1923). The results confirmed the two factor difference for roughness of awn already reported by other workers. The breeding behaviour of F_3 lines for resistance and susceptibility to rust was compared with awn character as in the following table, where 243 F_2 plants classified for their awn index were tested as F_3 lines for rust reaction.

		F_2 Awn Character.		
		Rough.	Intermediate Rough.	Smooth.
F_3 rust reaction.	Resistant or segregating.	171	43	11
	Susceptible ..	14	4	0
Total ..	.. .. ..	185	47	11

χ^2 for independence gave a P value 0·50–0·70 and the characters were considered to be independent.

Similar classifications were made for other characters and none of them revealed any association with leaf rust resistance. The following table summarizes the P values that were obtained (Table 7):

TABLE 7.

Characters.	Number of Plants.	χ^2.	P.
Roughness of awn and rust resistance	243	0·937	0·50–0·70
Rachilla hairs and rust resistance ..	243	0·044	0·70–0·80
Powdery mildew and rust resistance	237	3·201	0·50–0·70
Roughness of awn and rachilla hairs	263	18·680	<0·001

The association between roughness of awn and rachilla hair length was confirmed but as the result was based on less than 300 plants, no linkage values are given.

Crosses with Other Varieties.

The parents used in this study have been crossed with several other leaf rust resistant varieties to determine any relationship in the resistance to this disease. In the crosses used although the seed setting was good, grain mites lowered the viability of the crossed grain between the time of harvesting and sowing and hence the F_2 populations are small. In spite of the fact that they are too small for final conclusions to be drawn, they are reported here as an indication of the type of result obtained (Table 8).

TABLE 8.

Parents.	Number of Plants.	Classification.
Minn. II 21.15 × Virginia Hooded C.I. 2290	46	All resistant.
Minn. II 21.15 × *H. distichon rimpaui typica*	228	,, ,,
Minn. II 21.15 × Featherston	101	,, ,,
Minn. II 21.15 × Oderbrucker C.I. 940	460	,, ,,
Minn. II 21.15 × Hooded Spring C.I. 716	188	,, ,,
Minn. II 21.15 × Horsford C.I. 507	68	,, ,,
Virginia Hooded C.I. 2290 × Minn. II 21.15	166	,, ,,
Minn. 184 Manchuria × Minn. II 21.15	701	,, ,,
H. vulgare aethiops typica × Minn. II 21.15	47	,, ,,
Featherston × Minn. II 21.15	308	,, ,,
Oderbrucker C.I. 940 × Minn. II 21.15	486	,, ,,
Brachytic chlorina rust resistant × *H. distichon rimpaui typica*	424	,, ,,
H. distichon rimpaui typica × Brachytic chlorina rust resistant	118	,, ,,
H. vulgare aethiops typica C.I. 2208 × Virginia Hooded C.I. 2290	201	,, ,,

	Number of Plants.	R.	M. R.	S.
H. vulgare aethiops typica C.I. 2208 × No. 22	54	42	8	4
Virginia Hooded C.I. 2209 × No. 22	55	38	14	3
Featherston × No. 22	52	41	7	4
Oderbrucker C.I. 940 × No. 22	52	35	16	1
No. 22 × Virginia Hooded C.I. 2290	62	44	15	3
No. 22 × Featherston	56	43	11	2
No. 22 × Hooded Spring C.I. 716	60	46	10	4

R., resistant; M.R., moderately resistant; S., susceptible.

Certain of these crosses were made without knowing the genetic make-up of the parents for rust resistance. Despite this and despite the small populations that were tested, the results obtained to date indicate that there are probably not a great number of loci involved in conveying resistance to *P. anomala*. All the varieties that give a reaction like Minn. II 21.15 failed to give any segregation when crossed with it. No. 22, on the other hand, obviously had a different type of resistance and gave segregation in all cases where a cross was made with a variety showing the Minn. II 21.15 type of resistance. Observations are now being made on material in which No. 22 has been crossed with other varieties having a similar type of resistance.

SUMMARY.

Several collections of barley leaf rust have been shown to be similar, but unlike any of the 30 races described from Europe. Using one of these collections on F_1, F_2 and F_3 material of a cross II 21.15 (Smooth Awn × Manchuria) × No. 22, it was found that the two single factors possessed by these parents are not allelic and are inherited independently. The factor in II 21.15 (Smooth Awn × Manchuria) has been called Pa_1 and that in No. 22 Pa_2. When Smooth Awn × Manchuria was crossed with a number of varieties having a similar type of resistance, no segregation occurred. It appears from preliminary observation that there are not many loci involved in giving resistance to *P. anomala*.

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